

IT IS CLAIMED THAT:

1. A method for identifying a gene function in a plant comprising a conditional lethal mutation in a gene comprising the steps of:

- 5 (a) growing one or more plants under first permissive conditions to produce a group of plants;
- (b) growing a first set of the plants produced in step (a) under one or more restrictive conditions to determine the presence of a conditional lethal mutation;
- 10 (c) selecting one or more plants from step (b) that are sensitive to said restrictive conditions;
- (d) growing a second set of the plants, which are produced in step (a) and genetically identical to those selected in step (c), under second permissive conditions to determine the growth requirement of plants having the conditional lethal mutation;
- 15 (e) growing a third set of the plants, which are produced in step (a) and genetically identical to those selected in step (c), under said restrictive conditions, and complementing a mutated gene of said selected plants by transfecting them with a viral vector containing an unmutated copy of a mutated gene, thereby identifying a gene function in a plant comprising a
- 20 conditional lethal mutation in a gene.

2. A method for identifying a gene function in a plant which comprises a conditional lethal mutation in a gene, comprising:

- 25 (a) growing one or more plants under first permissive conditions;
- (b) growing a set of plants produced in step (a) under one or more restrictive conditions;
- (c) selecting one or more plants from step (b) that are sensitive to the restrictive condition;

- (d) growing a set of plants selected in step (c) under a variety of permissive conditions;
- (e) growing a set of plants selected in step (c) under a restrictive condition and complementing a mutated gene of the plants by transfecting the plants with a viral vector containing an unmutated copy of the mutated gene.
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3. The method of Claim 1 or 2, further comprising after step (e), the step of (f) isolating from said viral vector a gene complementing said mutation.
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4. The method of Claim 3, further comprising after the step of isolating said gene, a step selected from the group consisting of: (i) identifying the function of said gene, (ii) identifying the product expressed by said gene, and (iii) sequencing said gene.
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5. The method of Claim 1 or 2, wherein the first permissive conditions include a complete growth medium for the plant tissue, plant cell or plant organ.
6. The method of Claim 1 or 2, wherein the first permissive conditions include a growth medium at low osmotic strength.
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7. The method of Claim 1 or 2, wherein the first permissive conditions include a temperature between about 5 and 15°C below the optimal growth temperature for a wild type uninfected plant.
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8. The method of Claim 1 or 2, wherein the restrictive conditions include a temperature between the optimal growth temperature for the organism and at least about 15°C above the optimal growth temperature for the organism.
9. The method of Claim 1 or 2, wherein the second permissive conditions are
- 30 substantially the same as the first permissive conditions.

10. The method of Claim 1 or 2, wherein the plant cells in growing step (a) are replica plated plant cells on plant leaf disks.
- 5 11. The method of Claim 1 or 2, wherein the period of time in step (c) is equivalent to at least one growth cycle.
12. The method of Claim 1 or 2, wherein the plants from step (a) are selected from the group consisting of monocotyledons and dicotyledons.
- 10 13. The method of Claim 1 or 2, wherein the plants from step (a) have been mutagenized by insertion mutagenesis with T-DNA or transposon nucleic acid sequences.
- 15 14. The method of Claim 13, wherein the plants have been mutagenized with a mutagen selected from the group consisting of nucleic acid alkylating agents, intercalating agents, ionizing radiation, heat, and sound.
- 20 15. The method of Claim 14, wherein said alkylating and intercalating agents are selected from the group consisting of methanesulfonate, methyl methanesulfonate, methylnitrosoguanidine, 4-nitroquinoline-1-oxide, 2-aminopurine, 5-bromouracil, ICR 191 and other acridine derivatives, ethidium bromide, nitrous acid, and N-methyl-N'-nitroso-N-nitroguanidine.
- 25 16. The method according to Claim 1, wherein said plant is a transgenic plant.
17. The method according to Claim 1, wherein said plant is *Nicotiana benthamiana*, *Nicotiana tabacum* or *Arabidopsis thaliana*.

18. The method according to Claim 1, wherein said viral vector is derived from a single-stranded plus sense plant RNA virus.

19. The method according to Claim 18, wherein said viral vector is derived from a tobacco mosaic virus, tomato mosaic virus, or ribgrass mosaic virus.

20. A method for identifying a gene function in a plant carrying a conditional lethal mutation in a gene, comprising:

- (a) crossing to itself, a plant that is heterozygous for a conditional lethal mutation to make a homozygous mutant plant; and
- (a) growing the plant from step (a) under a restrictive condition and complementing a mutated gene of the plant by transfecting it with a viral vector containing an unmutated copy of the mutated gene.